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CDKAL1 and *HHEX* are associated with type-2 diabetes-related traits among Yup'ik people

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Abstract

Background: Genome-wide association studies (GWAS) have identified single nucleotide polymorphisms (SNPs) associated with type-2 diabetes (T2D), mainly among individuals of European ancestry. We examined the frequency of these SNPs and their association with T2D-related traits in an Alaska Native study population with a historically low prevalence of T2D. We also investigated whether dietary characteristics that may protect against T2D, such as n-3 polyunsaturated fatty acid (n-3 PUFA) intake, modify these associations.

Methods: In 1,144 Yup'ik people, we examined 17 SNPs repeatedly identified in GWAS for individual and cumulative associations with T2D-related traits. Cumulative associations were evaluated using a genetic risk score (GRS) calculated by summing risk alleles. Associations were tested for interactions with sex, BMI, and n-3 PUFA intake.

Results: The rs7754840 SNP in *CDKAL1* is significantly associated with HbA_{1c} (p=0.00091). The rs5015480 SNP near *HHEX* is significantly associated (in opposite direction to that in

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Significant findings of the study: We examine and compare the frequency of T2D-associated SNPs in Yup'ik people. SNPs in CDKAL1 and HHEX are found to be associated with glycemic traits in this sample. Associations are not modified by BMI or dietary n-3 PUFA intake.

What this study adds: This is the first study to examine the frequency of T2D SNPs in an Alaska Native population, which has a historically low prevalence of T2D. We examine associations between SNPs and glycemic traits, and interactions with BMI and n-3 PUFA intake.

DISCLOSURE
The authors have no potential conflict of interest to report.

Europeans) with a combined fasting glucose (FG) and HbA_{1c} measure (p=0.00046) and with HOMA-B (p=0.0014). The GRS is significantly associated with FG and combined FG & HbA_{1c} only when the *HHEX* SNP is dropped from the GRS. Associations are not modified by BMI or n-3 PUFA intake.

Conclusion: Our results highlight the potential importance of *CDKAL1* and *HHEX* in glucose homeostasis in this Alaska Native population with a low prevalence of T2D, and suggest that these loci should be examined in greater detail in this population.

Keywords

Alaska Native; CANHR; type-2 diabetes SNPs; glycemic traits

INTRODUCTION

Type-2 diabetes (T2D) is a major health concern, often disproportionately affecting Nativfe American populations.[1–3] Prior to the 1980s, epidemiological studies demonstrated that T2D among Alaska Native people was a rare condition.[4] Despite data suggesting that the prevalence of T2D is increasing among coastal Alaska Native communities, Yup'ik people from Southwest Alaska continue to demonstrate a T2D prevalence (4 to 6%) that is lower compared to the overall US population (8%), and may be independent of obesity status.[1,2,5,6]The mechanisms that allow Yup'ik people to carry excess body fat without developing T2D at the same rate as other populations may involve dietary and genetic factors.

Several studies suggest that genetic factors partially underlie T2D and related traits.[7,8] Genome-wide association studies (GWAS) for T2D and related traits have been performed mainly among individuals of European descent.[9–11] Subsequent studies have found evidence of consistent associations of these SNPs across populations[12–14], although associations at some loci, such as *TCF7L2*, do not appear to be generalizable.[15]Currently, little is known regarding the frequency of these SNPs among Alaska Native people, and whether they are associated with T2D traits.

Epidemiologic studies have reported a lower prevalence of impaired glucose tolerance and T2D in populations consuming large amounts of n-3 PUFAs found mainly in fish and marine mammals.[16] Yup'ik people have an unusually high variability in n-3 PUFA intake, because fish and marine mammals are an important part of the traditional diet, but adherence to this diet is highly variable.[17,18] Consumption of n- PUFAs may contribute to the low prevalence of T2D in this group,[19] and genetic factors may mediate this association.

Understanding the genetic mechanisms and dietary interactions underlying T2D pathophysiology will require knowledge regarding how loci identified in T2D GWAS are associated with specific T2D-related quantitative traits. Previous studies suggest considerable overlap of loci associated with T2D and related quantitative traits,[10,20,21] and that distinct genetic factors underlie each of these quantitative traits.[22,23]

In this study, we determine the extent to which 17 T2D-associated SNPs identified in the first wave of GWAS and confirmed in recent GWAS meta-analyses, are polymorphic, are associated with glycemic traits , and interact with n-3 PUFA intakein a sample of 1,144 Yup'ik people from a population with a historically low prevalence of T2D..

METHODS

Study sample

Through the Center for Alaska Native Health Research (CANHR), Yup'ik participants were recruited in 11 southwest Alaska communities to participate in a study examining genetic, behavioral, and dietary factors underlying metabolic traits. . The sample used here consists of 1,144 non-pregnant self-identified Yup'ik people between the ages of 14 and 94. Summary statistics regarding family structure among participants were calculated using PEDINFO in the Statistical Analysis for Genetic Epidemiology (S.A.G.E., 2009) software. The sample consists of 49 founders, 969 non-founders, and 123 singletons. There were 195 pedigrees with a mean size of 11.44 individuals (range, 1-894) and 696 sibships with a mean size of 1.39 individuals (range, 1-9). The age distribution in this study resembles the age distribution among eligible participants, according to the 2000 US Census data. Informed consent was obtained from all participants using protocols approved by the University of Alaska Institutional Review Board, the National and Alaska Area Indian Health Service Institutional Review Boards, and the Yukon Kuskokwim Health Corporation Human Studies Committee.

Phenotypic and dietary measurements

Height and weight measurements were obtained by trained staff using protocols from the NHANES (National Health and Nutrition Examination Survey) III Anthropometric Procedures Manual,[24] as previously described.[5] Fasting insulin (FI) was assayed with radioimmunoassay kit using an I^{125} –iodinated insulin tracer, anti-human insulin specific antibody, and human insulin standards from Linco Research where the intra- and inter-assay variations were 5.8% and 10.2%, respectively. Fasting blood glucose (FG) was measured on a Cholestech LDX analyzer, and glycosylated hemoglobin (HbA_{1c}) was measured on a Bayer HbA_{1C} DCA 2000+ analyzer (Bayer AG, Leverkusen, Germany). We calculated HOMA-IR (homeostatic model assessment of insulin resistance) as the product of FG and FI divided by 22.5, and HOMA-B (homeostatic model assessment of beta-cell function) as (FI * 360) / (FG – 63).[25] Finally, we defined a continuous variable called 'FG & HbA_{1c}' based on previous studies indicating that combined information from FG and HbA_{1c} is a reliable predictor of developing T2D.[26,27] The FG & HbA_{1c} variable was calculated by first standardizing each variable to have a standard deviation of 1, then using the mean of the two standardized variables. Data on diabetes-related medications (metformin, glyburide, pioglitazone) were obtained through self-report at the time participants were enrolled in the study, and extracted from the Yukon Kuskokwim Health Corporation medical records by a trained nurse. Long-chain n-3 PUFA intake was assessed using the nitrogen stable isotope ratio ($^{15}N/^{14}N$) of red blood cells, as previously described.[18,28]

Genotypes

Genes selected for this study included those identified in the first wave of GWAS for T2D,[29–32] and also replicated in a meta-analysis.[10] Specific SNPs within or near each gene were selected based on which has been associated with T2D in more than one GWAS, as of June, 2011. Genotyping was conducted at the Broad Institute (Cambridge, MA) using a Sequenom iPLEX platform.[33] The *FTO* SNP (rs9939609) was previously investigated for association with body composition traits in this sample.[34] We excluded SNPs with> 10% missing frequency or a minor allele frequency < 1%. Allele frequencies and deviations from Hardy-Weinberg equilibrium were evaluated using MENDEL software,[35] accounting for family structure. We compared allele frequencies in this sample to those in five HapMap reference populations.[36]

Genetic risk score

A genetic risk score (GRS) was calculated for all individuals with at least six non-missing genotypes for the fourteen SNPs considered. Risk alleles were defined as those which are positively associated with T2D risk in GWAS. The sum of risk alleles was divided by the number of non-missing genotypes, to account for missing genotypes. We also used a weighted GRS, with weights corresponding to published T2D odds ratios.[10]

Statistical analyses

Linear models were fit to each phenotype using the following covariates: age, sex, BMI, medication use, and community location. We ran analyses with and without BMI as a covariate to evaluate the extent to which significant associations with T2D-related traits were mediated by obesity. The community location variable is based on the second principal component from a principal components analysis (PCA) of 4,108 autosomal markers, as previously described,[28] and corresponds to the proximity of each community to the coast. It is included as a covariate in the analyses to control for population stratification. The first component from this PCA had no obvious systematic structure, as previously described.[37] As a sensitivity analysis, we examined the effect of controlling for the first PC on our main findings.. The distributions of residuals from linear models were examined for normality, and Box-Cox transformations [38] and extreme outlier removal were implemented for each phenotype that deviated from the normality assumption.

We accounted for within-pedigree correlation using the linear mixed effects model (LME) implemented in the 'lmekin' function in the *coxme* package (http://cran.r-project.org/web/packages/coxme/index.html) [39] to test the association between each phenotype and each SNP, using an additive genetic model. We tested the cumulative association of these SNPs with each phenotype using the GRS, including the same covariates listed above . We also examined interactions with sex, BMI, and n-3 PUFA intake. We used a Bonferroni correction for multiple testing (14 tests) in the single SNP analyses ($\alpha = 3.57 \times 10^{-3}$).

RESULTS

Descriptive Statistics

The sample consists of 540 males and 604 females. Mean phenotypic values for the entire sample and for each sex are listed in Table 1. Females have significantly higher BMI, FG, FI, HOMA-IR, and HOMA-B, and significantly lower FG. Thirteen individuals reported taking T2D-related medication. After excluding outliers (FG>215, HbA_{1c}>9) and individuals taking medication, we find that 0.97% of individuals have a FG above 125 mg/dL, 1.3% of individuals have an HbA_{1c} equal or greater than 6.5%, 20% have a FG between 100 and 125 mg/dL, and 24% of individuals have an HbA_{1c} between 5.7 and 6.4%.

Two of the 17 SNPs (rs10923931-*NOTCH2* and rs7578597-*THADA*) had an extremely low MAF (<0.01), and one SNP had a missing rate > 10% (rs1801282 – *PPARg*), thus precluding them from analyses.. Our analysis therefore included a total of 14 SNPs. The frequency of the risk allele for each SNP in this sample and in selected HapMap populations is listed in Table 2. The allele frequencies in Yup'ik people do not differ appreciably from that of geographically close populations (e.g. CHB). All SNPs are in Hardy Weinberg equilibrium after correction for multiple testing ($p > 0.0036$).

Association of individual SNPs and n-3 PUFA with phenotypes

We find that n-3 PUFA intake is positively and significantly associated with HbA_{1c} ($p=0.02$), after adjusting for covariates.

We find that 10 of the 14 SNPs are associated with increased HbA_{1c} and FI in a direction consistent with GWAS findings for T2D. 9 out of 14 SNPs are associated with FG, FI, HOMA-IR, and the combined FG & HbA_{1c} measure in a direction consistent with T2D GWAS. 8 out of 14 T2D GWAS risk alleles are associated with a decrease in HOMA-B (see Table 3).

The only statistically significant SNP associations, after correction for multiple testing, are between rs7754840 (*CDKAL1*) and HbA_{1c} ($p=9.1 \times 10^{-4}$), and between rs5015480 (*HHEX*) and the combined FG & HbA_{1c} measure ($p=4.6 \times 10^{-4}$), as well as with HOMA-B ($p=1.4 \times 10^{-3}$) (see Table 3). These associations do not appear to be driven principally by BMI, as the strength of association differs minimally upon adjusting for BMI (see Table 3). Controlling for the 1st PC very slightly attenuated the strength of both associations, but results were still statistically significant. Interestingly, the associations with the SNP in *HHEX* are in the opposite direction compared to GWAS findings. The present analysis revealed that the T allele is associated with higher trait values (and lower HOMA-B), while in GWAS, the C allele is associated with greater T2D risk. Variation at rs7754840 explains approximately 0.8% of variation in HbA_{1c}, and variation at rs5015480 explains approximately 0.8% of variation in FG & HbA_{1c} and HOMA-B. The association between the *MTNR1B* SNP (rs10830963) and HOMA-B is marginally significant ($p=6.4 \times 10^{-3}$).

We found no evidence that associations were significantly modified by BMI, sex, or n-3 PUFA intake (see Supplementary Table 1-3). Based on a previous study that found that the associations between both *CDKAL1* and *HHEX* variants and T2D risk were stronger in females than males [40], we examined associations stratified by sex. The association between rs7754840 (*CDKAL1*) and HbA_{1c} is significant in females (p=0.00053), but not in males (p=0.35). The p-value for the interaction of this SNP and sex is nominally significant (p=0.013).

Association of T2D GRS with phenotypes

The T2D GRS is positively associated with all phenotypes, although none of the associations are statistically significant (see Table 3). Since the associations with the *HHEX* SNP were in the opposite direction in this sample, compared to GWAS, we computed a GRS excluding this SNP. We find that this GRS is positively and significantly associated with FG & HbA_{1c} (p=0.014) and with FG (p=0.021). The proportion of variance in these traits explained by this modified GRS is 1.2% or less. Results did not improve or change significantly with a weighted GRS. Using a risk score without *CDKAL1* (and without *HHEX*), the p-value for association increases to 0.11 for FG & HbA_{1c} and to 0.055 for FG.

We examined risk scores in which only nominally significant ($p<0.05$) SNPs in a consistent direction are included in the score. For FG, a risk score comprised of *IGF2BP2* and *MTNR1B* is nearly significantly associated with fasting glucose (p=0.05). For the other traits, there are either no nominally significant associations with any SNPs, or the only significant SNP (in a consistent direction) is *CDKAL1* or *MTNR1B*.

Using the GRS without the *HHEX* SNP, we find no evidencethat associations between the GRS and FG and FG & HbA_{1c} are modified by sex, BMI, or n-3 PUFA intake.

DISCUSSION

We examined whether SNPs identified in T2D GWAS among individuals of European descent are associated with T2D-related traits among Yup'ik people who have a historically low prevalence of T2D. The prevalence of prediabetes (defined as HbA_{1c} greater than 5.6

but less than or equal to 6.5, or FG greater than 99 but less than 126) in this sample of Yup'ik people was relatively low (22%), compared to 35% in the adult US population.[41]

Variation in *CDKAL1* (rs7754840) was significantly associated with HbA_{1c}. This SNP is located within intron 5 of the *CDKAL1* (CDK5 regulatory subunit associated protein 1-like 1) gene.*CDKAL1* variants have been found to be associated with reduced beta-cell glucose sensitivity and reduced insulin secretion.[42,43] A GWAS for HbA_{1c} among normoglycemic Koreans identified *CDKAL1* as most strongly associated with HbA_{1c},[44] and a study among healthy Japanese men found an association between *CDKAL1* and HbA_{1c}.[45] As previously reported in a cohort of Korean individuals,[40] we find that this association is stronger among females than males.

Variation near *HHEX* (rs5015480) was significantly associated with FG & HbA_{1c} and HOMA-B. This SNPis located upstream of the *HHEX* (hematopoietically expressed homeobox) gene which has been found to be involved in ventral pancreatic development by controlling the proliferation rate of endodermal cells.[46] This SNP was previously found to be associated with decreasedinsulin response and beta-cell glucose sensitivity.[47] Both *CDKAL1* and *HHEX* variants were strongly associated with T2D and insulin secretion in several Southeast Asian cohorts and among Pima Indians, respectively.[54,55] Our finding that the risk allele for the HHEX SNP as identified in GWAS corresponds to the "protective" allele in this sample is not uncommon in genetic studies,[48] and may be attributed to population-unique linkage-disequilibrium patterns, or that the identified SNP is likely not the causal variant. The marginally significant association between rs10830963 in *MTNR1B* (melatonin receptor 1B) and HOMA-B is consistent with previous findings showing that variation in *MTNR1B* is associated with HOMA-B and other insulin secretion measures.[49–51]*MTNR1B* is expressed in pancreatic islets, and has been implicated in the regulation of circadian rhythms and insulin secretion.[52,53]

Finally, our analysis revealed the GRS without the *HHEX* SNP was significantly associated with FG and FG & HbA1c, although these associations appear to be driven in large part by the associations with *CDKAL1*.

The strengths of this study include detailed phenotypic measurements in a population with a historically low prevalence of T2D and highly variable PUFA intake. However, there are limitations that should be noted. Measures of HbA_{1c} can partly reflect other hematological traits such as hemoglobinopathies,[56] therefore potentially confounding associations. Secondly, we have no estimate of total caloric or fat intake in the entire sample to include as a covariate in associations involving n-3 PUFA intake, making it difficult to interpret our finding of a positive association between n-3 PUFA intake and HbA_{1c}. For a subset of participants (n=515), total energy and percentage fat intake (% of total calories) data from 24 Hour Recall was available. Within this subset, the association between n-3 PUFA intake and HbA_{1c} is unchanged upon including total energy intake as a covariate. However, upon including % fat intake as a covariate, the association is no longer statistically significant (data not shown), suggesting that additional work is needed to understand the relationship between n-3 PUFA intake and T2D traits.

It should be noted that the discovery of additional T2D susceptibility variants has continued in recent years, with the total number of confirmed SNPs up to approximately 65.[20] SNPs included in our study were identified in the first wave of GWAS meta-analysis and may be those with relatively larger effect sizes. Nevertheless, we acknowledge that there are likely hundreds or thousands of variants implicated inT2D.Our results show that Yup'ik study participants have a low prevalence of prediabetes relative to other populations, and that *CDKAL1* and *HHEX* variants are associated with FG and HbA1c. Future studies should fully

characterize variation in the regions surrounding these genes in an effort to elucidate how the molecular mechanisms underlying these phenotypes are mediated by genetic variation and interact with lifestyle and behavioral factors.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Table 1

Descriptive Statistics (s.d.: standard deviation, * denotes statistically significant difference between males and females)

	Mean (s.d.)	Males (n=540)	Females (n=604)
Age	36.85 (17.35)	35.97 (17.34)	37.63 (17.33)
BMI (kg/m^2) *	27.40 (6.08)	25.93 (4.82)	28.7 (6.76)
Fasting glucose (mg/dL) *	93.14 (10.98)	94.30 (11.20)	92.10 (10.69)
HbA_{1c} (%)	5.46 (0.39)	5.49 (0.35)	5.43 (0.42)
Fasting insulin (mU/L) *	14.62 (8.08)	13.49 (7.33)	15.58 (8.55)
HOMA-IR *	3.43 (2.27)	3.21 (2.21)	3.62 (2.30)
HOMA-B *	189.8(117.7)	167.1 (96.7)	209.0 (130.0)

Table 2

T2D risk SNPs, nearest gene, and allele frequencies.

				Risk allele frequency in reference populations				
SNP	**Gene**	**Risk allele**	**Risk allele frequency in sample**	**YRI**	**CEU**	**CHB**	**JPT**	**MXL**
rs4607103	*ADAMTS9*	C	0.60	0.69	0.79	0.57	0.60	0.67
rs4402960	*IGF2BP2*	T	0.28	0.53	0.31	0.27	0.33	0.21
rs7754840	*CDKAL1*	C	0.44	0.68	0.32	0.43	0.41	0.30
rs864745	*JAZF1*	T	0.60	0.78	0.49	0.77	0.82	0.64
rs13266634	*SLC30A8*	C	0.61	0.93	0.76	0.55	0.55	0.78
rs10811661	*CDKN2A*	T	0.79	0.97	0.82	0.58	0.54	0.87
rs12779790	*CDC123*	G	0.11	0.11	0.25	0.19	0.11	0.12
rs5015480	*HHEX*	C	0.35	0.62	0.57	0.20	0.19	0.51
rs7901695	*TCF7L2*	C	0.08	0.40	0.34	0.03	0.03	0.24
rs2237892	*KCNQ1*	C	0.59	0.90	0.92	0.65	0.62	0.76
rs5215	*KCNJ11*	C	0.40	0.02	0.37	0.38	0.35	0.42
rs7961581	*TSPAN8*	C	0.08	0.18	0.26	0.18	0.24	0.14
rs10830963	*MTNR1B*	G	0.32	0.03	0.26	0.45	0.46	0.22
rs9939609	*FTO*	A	0.16	0.50	0.46	0.15	0.19	0.23

Table 3: Association of T2D SNPs with T2D-related quantitative traits with and without adjustment for BMI. Reference allele refers to the risk allele as determined by GWAS in Europeans.

SNP	Gene	Ref. & risk Allele	FG	FI	HbA_{1c}	FG & HbA_{1c}	HOMA-IR	HOMA-B
rs4607103	*ADAMTS9*	C	−0.091,p=0.57	0.21, p=0.25	0.06, p=0.75	6×10^{-5}, p=1	0.01, p=0.38	0.03, p=0.16
	w/out BMI		−0.096, p=0.56	0.28, p=0.17	0.03, p=0.88	−0.0003, p=0.99	0.02, p=0.27	0.03, p=0.11
rs4402960	*IGF2BP2*	T	0.429, p=0.021	0.1, p=0.65	−0.09, p=0.68	0.04, p=0.22	0.03, p=0.13	−0.03, p=0.25
	w/out BMI		0.36, p=0.059	−0.07, p=0.78	−0.19, p=0.42	0.03, p=0.49	0.01, p=0.51	−0.04, p=0.12
rs7754840	*CDKAL1*	C	0.222, p=0.18	0.13, p=0.48	0.68, p=0.00091	0.08, p=0.012	0.01, p=0.37	−0.01, p=0.59
	w/out BMI		0.203, p=0.23	0.14, p=0.52	0.72, p=0.00074	0.08, p=0.012	0.02, p=0.4	−0.01, p=0.62
rs864745	*JAZF1*	T	0.179, p=0.28	0.06, p=0.74	0.08, p=0.7	0.03, p=0.41	0.01, p=0.59	−0.01, p=0.49
	w/out BMI		0.204, p=0.23	0.05, p=0.82	0.05, p=0.8	0.02, p=0.46	0.01, p=0.67	−0.02, p=0.45
rs13266634	*SLC30A8*	C	0.123, p=0.45	0.16, p=0.4	0.05, p=0.81	0.02, p=0.51	0.01, p=0.66	0.02, p=0.34
	w/out BMI		0.133, p=0.43	0.23, p=0.27	0.09, p=0.65	0.03, p=0.39	0.01, p=0.47	0.02, p=0.28
rs10811661	*CDKN2A*	T	−0.067, p=0.73	−0.25, p=0.27	0.04, p=0.89	0.01, p=0.77	−0.01, p=0.63	−0.03, p=0.32
	w/out BMI		−0.07, p=0.73	−0.21, p=0.41	0.04, p=0.88	0.02, p=0.7	−0.01, p=0.8	−0.02, p=0.37
rs12779790	*CDC123*	G	0.374, p=0.17	0.21, p=0.49	0.05, p=0.87	0.07, p=0.21	0.03, p=0.32	−0.005, p=0.9
	w/out BMI		0.341, p=0.22	0.07, p=0.84	0.07, p=0.85	0.07, p=0.23	0.02, p=0.58	−0.01, p=0.7
rs5015480	*HHEX*	C	−0.496, p=0.0043	0.21, p=0.29	−0.61, p=0.0046	−0.12, p=0.00046	−0.01, p=0.49	0.07, p=0.0014
	w/out BMI		−0.395, p=0.027	0.35, p=0.11	−0.49, p=0.03	−0.1, p=0.0066	0.002, p=0.91	0.08, p=0.00064
rs7901695	*TCF7L2*	C	−0.263, p=0.42	0.28, p=0.47	−0.69, p=0.094	−0.1, p=0.11	0.004, p=0.9	0.04, p=0.34
	w/out BMI		−0.325, p=0.33	0.21, p=0.62	−0.71, p=0.097	−0.11, p=0.1	−0.001, p=0.98	0.04, p=0.41
rs2237897	*KCNQ1*	C	0.222, p=0.19	0.16, p=0.45	−0.15, p=0.5	0.03, p=0.39	0.02, p=0.23	−0.02, p=0.41
	w/out BMI		0.344, p=0.066	0.05, p=0.81	−0.2, p=0.4	0.02, p=0.63	0.01, p=0.51	−0.02, p=0.32
rs5215	*KCNJ11*	C	0.0425, p=0.8	−0.25, p=0.21	0.15, p=0.49	0.01, p=0.69	−0.01, p=0.5	−0.02, p=0.28
	w/out BMI		−0.043, p=0.8	−0.41, p=0.056	0.02, p=0.92	−0.01, p=0.85	−0.03, p=0.17	−0.03, p=0.13
rs10830963	*MTNR1B*	G	0.4234, p=0.018	−0.3, p=0.15	0.27, p=0.23	0.06, p=0.073	0.004, p=0.84	−0.06, p=0.0064
	w/out BMI		0.431, p=0.018	−0.41, p=0.073	0.15, p=0.51	0.05, p=0.19	−0.005, p=0.8	−0.07, p=0.0029
rs7961581	*TSPAN8*	C	−0.00083, p=1	−0.23, p=0.5	0.05, p=0.9	−0.01, p=0.82	−0.01, p=0.63	−0.02, p=0.65
	w/out BMI		0.077, p=0.81	−0.14, p=0.71	−0.03, p=0.94	−0.01, p=0.82	−0.01, p=0.84	−0.01, p=0.81
rs9939609	*FTO*	A	−0.272, p=0.22	8×10^{-5}, p=1	−0.04, p=0.88	−0.03, p=0.48	−0.01, p=0.7	0.03, p=0.33
	w/out BMI		−0.145, p=0.52	0.35, p=0.21	0.06, p=0.84	−0.002, p=0.96	0.02, p=0.4	0.05, p=0.086

Table 4

Association of T2D genetic risk score (GRS) with T2D-related quantitative traits. (p-values shown; statistically significant associations shown in bold)

	FG	**FI**	**HbA_{1c}**	**FG & HbA_{1c}**	**HOMA-IR**	**HOMA-B**
GRS	0.17	0.50	0.42	0.21	0.238	0.7
GRS without *HHEX*	**0.021**	0.75	0.08	**0.014**	0.148	0.15